

Development of Primary Secretary Ducts in the Stem of *Mangifera indica* L. (Anacardiaceae)

by

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Summary

The development of primary secretory ducts of stems of *Mangifera indica* L. has been studied with the aid of the electron microscope. The duct cavity has been found to be formed lysigenously. The primary ducts start to develop in the young leaf primordia. There the future epithelial cells still contain very large central vacuoles. These cells envelope a single file of cells which disintegrate and initiate the duct cavity. In the stem the duct cavity enlarges by lysis of epithelial cells and neighbouring cells become epithelial. In open ducts wall-remains of disintegrated cells are found attached to the active epithelial cells.

Introduction

Many early botanists with their excellent minds and with the aid of the then available equipment presented descriptions of the structure of cells and tissues, which, when examined many years later with the aid of the electron microscope, were usually found to be accurate. However, new embedding and sectioning techniques for light microscopy and the use of the electron microscope for anatomical investigations have in many cases added important new information to the understanding of developmental processes of cells and tissues.

The mode of initiation and development of cavities of internal secretory structures of plants has been dealt with mainly in early studies. Various investigators often held contradictory views as to whether cavities of specific ducts develop schizogenously, lysigenously or schizo-lysigenously (cf. Carr and Carr, 1970).

The ducts of the Anacardiaceae were examined by many researchers. The views on the manner of formation of the duct cavity in the different species and organs vary to a great extent (Müller, 1866-67; Sieck, 1895; Tschirch, 1900; Venning, 1948; Varghese and Pundir, 1964; Fahn and Evert, 1974). While studying the ducts of *Mangifera indica* and the way their cells produce and eliminate their secretory substance, we also tried to clarify the manner of duct cavity development in the different plant organs, with the aid of the electron microscope.

The present paper deals with the development of the primary shoot ducts.

Material and Methods

Stem portions from the apical region and from mature internodes of one-year-old saplings of mango (*Mangifera indica* L.) were used for examination. The saplings were grown in a greenhouse at the Hebrew University of Jerusalem.

For light microscopy hand cut sections were used.

For electron microscopy small portions of tissues including ducts were fixed in 6% glutaraldehyde for 2 hours, postfixed in OsO_4 2% for 2 hours (both in cacodylate buffer 0.1M pH 7.2), dehydrated in ethanol and embedded in Spurr's low viscosity embedding medium (Spurr, 1969). Sections were cut on LKB

ultratome III, stained with uranyl acetate and lead citrate, and examined with a Philips 300 electron microscope.

Observations

In the stems of mango (*Mangifera indica*) the primary secretory ducts occur in the phloem and pith (Plate 1, A-C). The mature duct consists, in cross section, of a cavity which is surrounded by a few concentric rows of cells which are more or less isodiametric or somewhat flattened and smaller than those of the neighbouring tissues. In the direction of the long axis of the duct they are elongated. No distinct intercellular spaces occur between these cells. The cells of the row neighbouring the cavity do not differ much from the cells of the other rows. They contain a large central vacuole and a thin layer of cytoplasm (Plate 2, A). In the duct cavity the secreted material is confined to its periphery. Most of the secreted material is exuded from the duct cavity during cutting of the stem and the fixation of the material.

The ducts start to differentiate in the shoot apex. In order to follow their development, successive sections, starting about 6 mm from the shoot tip and proceeding upwards, were examined. In the upper young region of the stem most of the cells neighbouring the duct cavity, the epithelial cells, are rich in cytoplasm and contain only a few very small vacuoles (Plate 2, B; Plate 4, A). The cytoplasm of these cells is dense, the ER is well developed. Parallel aligned ER elements have sometimes been seen to occupy a large part of the cytoplasm (Plate 4, A). The nuclei are relatively large. Plastids and mitochondria are numerous. Golgi bodies are occasionally observed in groups (Plate 3, A). Osmiophilic droplets are common and often in association with plastids and Golgi bodies (Plate 3, A; Plate 4, A, B). The process of secretion and sites of synthesis of the secretory substances in the cells will be treated in a separate article.

The duct cavity in the young portions of the stem is filled with electron dense secreted substances (Plate 4, B).

At the level of about 6 mm from the tip of the shoot apex single cells with a large vacuole and a very thin layer of cytoplasm are observed among the typical epithelial cells. Cell "a" in Plate 2, B gives the impression of joining the row of the densely stained epithelial cells after having divided from a cell situated behind them. In the gap where this cell is seen to join the epithelial cells, remnants of the back wall of an epithelial cell which has apparently undergone lysis can be seen. The region of the middle lamella between the wall of the joining cell and the remains of the wall of the disintegrated epithelial cell can clearly be observed.

In a section at a somewhat higher level (Plate 4, A), two small, narrow and deformed epithelial cells (d) with a dark disordered content are present. Between two unchanged epithelial cells a wide gap (g) can be seen in the same section. Part of the gap is occupied by electron dense material. One may assume that a cell which was in this place has disintegrated. Wall remains of the missing cell are seen attached to the neighbouring cell of the row surrounding the epithelial cells. In addition to outer cells joining the epithelial cells new cells are formed by anticlinal division of the epithelial ones. Between the cytoplasm of the epithelial cells and their walls, mostly in the region facing the duct cavity, large spaces occur containing vesicular and membranous structures, as well as very dark osmiophilic material (Plate 4, B). A thin layer of osmiophilic material often surrounds the entire cytoplasm.

In all sections, walls of the epithelial cells facing the cavity consist of two layers between which a distinct middle lamella can be seen. The middle lamella is swollen in many places (Plate 3, A, B). The layer closer to the cavity is irregularly fringed. This layer apparently represents remains of walls of the cells which have disintegrated.

The continuation of the stem ducts was followed in the leaf primordia situated less than 1 mm from the tip of the shoot apex. In the stem, before entering the primordia, the epithelial cells which are rich in cytoplasm were seen, in cross section, to surround a cell with a disintegrated protoplast (Plate 5A). In the leaf primordium itself the future epithelial cells contained large central vacuoles and thin layers of cytoplasm. As at the base of the leaf, at the site of the future duct cavity a single cell with a disintegrated protoplast was found in cross sections, at least up to one third of the height of the leaf primordium (Plate 5, B). In few places initial stages of disintegration were also observed in the cell wall.

Discussion

Diverse views of the mode of development of the duct cavity in the Anacardiaceae have been held by different authors. Engler (1896) dealing with the family, McNair (1918) who investigated *Rhus diversiloba* Torr. and Gray, and Fahn and Evert (1974) who investigated the secondary phloem ducts of *Rhus glabra* Thunb. stated that the ducts develop schizogenously. Sieck (1895), who worked on stems and fruits of *Anacardium occidentale* L. reported that the ducts originate schizogenously, but that continued development is lysigenous. Such a mode of duct development in Anacardiaceae was also reported by Tschirch (1900). Venning (1948) came to the conclusion that the manner of development of duct cavities varies among plant organs being of schizogenous origin in stems and leaves of *Schinus* and fruits of mango of schizo-lysigenous origin in stems and leaves of *Spondias* and mango and of lysigenous origin in floral organs (with the exception of the ovary) in mango. Varghese and Pundir (1964) reported that in the pseudocarp of *Anacardium occidentale* the ducts develop lysigenously.

The present electron microscopical investigation on the primary secretory ducts of the stem of mango showed that the cavities of these ducts initiate and develop lysigenously. The stem ducts initiate in the leaf primordia situated very close to the tip of the shoot apex. In the centre of the developing duct there is a single file of cells. The cells of this file disintegrate, initiating the development of the duct cavity.

In the stem, the duct cavity enlarges by lysis of epithelial cells, and neighbouring cells become epithelial. A proof for lysis of cells may be found in that remains of additional cell walls can be seen on the surface of the epithelial cell walls facing the duct cavity. Between these remains and the intact epithelial cell walls a middle lamella with occasional swellings can clearly be observed. It can thus be concluded that the lysis of the wall starts from inside the cell and progresses towards the middle lamella. This differs completely from the manner of wall lysis during schizogenous development of intercellular spaces of duct cavities (Fahn and Benajoun, in preparation).

Fractures occurring in the separation zone of leaves were also reported to be a result of the dissolution of the middle lamellar region of cell walls (Sexton and Hall, 1974).

The succession of wall lysis as seen in the mango ducts differs also from that reported by Tschirch (1889) for gum duct formation. There, according to Tschirch, the disintegration of walls starts in the primary walls and then proceeds in the secondary wall. The wall lysis in the cells of mango stem ducts differs therefore basically in mode of succession from that occurring in schizogenous processes and lysigenous formation of gum ducts.

Concurrently with the further process of cell lysis radial (anticlinal) divisions of epithelial cells may apparently take place. However, the initial stage of duct formation is strictly lysigenous.

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References

- CARR, D. J. & S. G. M. CARR, 1970. Oil glands and ducts in *Eucalyptus* l'Hérit. II. Development and structure of oil glands in the embryo. *Aust. J. Bot.* **18**: 191–212.
- ENGLER, A. 1896. Anacardiaceae. In: A. Engler and K. Prantl (ed.), *Die natürlichen Pflanzenfamilien* III, **5**.
- FAHN, A. & R. F. EVERT, 1974. Ultrastructure of the secretory ducts of *Rhus glabra* L., *Am. J. Bot.* **61**: 1–14.
- McNAIR, J. B. 1918. Secretory canals of *Rhus diversiloba*. *Bot. Gaz.* **65**: 268–273.
- MÜLLER, N. J. C. 1866–67. Untersuchungen über die Vertheilung der Harze, ätherischen Oele, Gummi und Gummiharze, und die Stellung der Sekretionsbehälter im Pflanzenkörper. *Jb. wiss. Bot.* **5**: 387–421.
- SEXTON, R. & J. L. HALL, 1974. Fine structure and cytochemistry of the abscission zone cells of *Phaseolus* leaves. I. Ultrastructural changes occurring during abscission. *Ann. Bot.* **38**: 849–854.
- SIECK, W. 1895. Die Schizolysigenen Sekretbehälter. *Jb. wiss. Bot.* **27**: 197–242.
- SPURR, A. R. 1969. A low-viscosity epoxy resin embedding medium for electron microscopy. *J. Ultrastruct. Res.* **26**: 31–43.
- TSCHIRCH, A. 1889. *Angewandte Pflanzenanatomie*, Band I. Urban & Schwarzenberg, Vienna.
- 1900. *Die Harze u.d. Harzbehälter*. Gebrüder Borntraeger, Leipzig.
- VARGHESE, T. M. & Y. P. S. PUNDIR, 1964. Anatomy of the pseudocarp in *Anacardium occidentale* L., *Proc. Indian Acad. Sci.* **59**: 252–258.
- VENNING, F. D. 1948. The ontogeny of the laticiferous canals in the Anacardiaceae. *Am. J. Bot.* **35**: 637–644.

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Plate 1. Micrographs of hand cut cross-sections of a stem, showing *A*, primary (phd) and secondary (sd) phloem ducts, $\times 120$; *B*, one primary phloem duct, $\times 300$; *C*, a pith duct (pd), $\times 300$.